STATEMENT OF PROJECT OBJECTIVES

Health Effects of Sub-chronic Inhalation of Simulated Downwind Coal Combustion Emissions
DE-FC26-05NT42304

A. SCOPE OF WORK

The work will encompass: 1) establishing a laboratory-generated exposure atmosphere containing key particulate and gaseous species in ratios considered appropriate by atmospheric and combustion scientists; 2) conducting subchronic (daily up to 6 months) repeated inhalation exposures of rodents to four graded dilutions of the atmosphere producing dose-response data down to environmentally-relevant concentrations; 3) measuring a spectrum of health outcomes and indications of mechanisms in the respiratory and cardiovascular systems that span multiple key public health concerns; 4) evaluating and publishing results demonstrating the presence or absence of health hazards and the nature of the dose-response curve, including evidence of thresholds; and 5) comparing the biological effects of coal combustion emissions to effects of other source emissions measured using the identical experimental protocol.

Phase I will develop the exposure atmosphere, including setting up a drop-tube furnace, developing the emissions modification system, establishing operating conditions necessary to achieve an exposure atmosphere meeting consensus criteria developed in an expert workshop, and comparing results from Powder River Basin sub-bituminous (PRB) and Low Sulfur Southern Appalachian bituminous (LSSA) coals.

Phase II will conduct the toxicological study using one coal type, including exposures to four dilutions of the atmosphere for times ranging from a few days to six months (depending on the health endpoint), conducting health assays according to the protocol used successfully in studies of diesel emissions, hardwood smoke, gasoline emissions, and street dust by the National Environmental Respiratory Center (NERC, www.nercenter.org) program at the Lovelace Respiratory Research Institute (LRRI), analyzing exposure concentration-response relationships, comparing results to those from studies of the other source emissions, and publishing the results and analyses.

B. TASKS TO BE PERFORMED

Phase I: Development of the Exposure Atmosphere

Task 1: Assemble Drop-Tube Furnace and Emissions Dilution/Modification System

A laboratory-scale combustion system based on a drop-tube furnace will be assembled. System components will be purchased. PRB and LSSA coals will be obtained in quantities necessary to complete the toxicology study, processed to a particle size suitable for use in the furnace, mixed well, and stored under nitrogen in sealed containers. The system will be assembled, revised as necessary to achieve proper function, and tested for safety. Standard operating procedures will be developed.

Task 2: Conduct Iterative Generation/Modification Trials Using PRB Coal

The initial assumption, based on workshop recommendations, will be that PRB coal will be used; thus, trials will first be conducted to determine the conditions necessary for achieving the desired exposure atmosphere using PRB coal. The composition of the exposure atmosphere in an animal exposure chamber (without animals) will be analyzed in detail, including detailed physical-chemical characterization of particulate, vapor, and gas phases. Iterative generation trials will be conducted to modify generation conditions and the addition of materials to the furnace effluent in order to develop an exposure atmosphere best matching the target composition. The results using PRB coal will be summarized.

Task 3: Conduct Generation Trials Using LSSA Coal

Once generation conditions have been optimized for PRB coal (Task 2), trials will be conducted using LSSA coal. The composition of the exposure atmosphere will initially be determined using the same operating parameters as used for PRB coal. If the exposure atmosphere is significantly different with LSSA coal, it will then be determined how closely it is possible to achieve the target composition using LSSA coal and different operating parameters. Results using LSSA coal will be summarized.

Task 4: Summarize Phase I Experience and Proposed Strategy for Phase II Exposures

All experience and results developed during the preceding tasks will be summarized in a written report. This report will provide a basis for a go/no go decision about proceeding with Phase II, and if so, the coal type and operating parameters to be used in Phase II.

Phase II: Conduct of the Toxicology Study

This phase will use an approach identical (with the exception of the exposure atmosphere) to that employed in the NERC program to study other source emissions. This strategy not only ensures that the study will be conducted using proven, well-standardized methods, but will also provide the unique opportunity for direct comparisons of the health hazards of coal emissions to those of other common mobile and stationary source emissions. The protocol is described in detail in Section III.B below. The experimental design and selection of health assays follow consensus recommendations of peer workshops of experimental design and health experts held in 1999 at LRRI at the inception of the NERC program, and updated since as dictated by evolving knowledge and experience.

Task 1: Set Up and Test Multi-Level Inhalation Exposure System

The system for generating the atmospheres developed and characterized in Phase I will be expanded to accommodate the multiple exposure levels and large number of animals required for the Phase II toxicology study. The system and operating controls will be constructed. The performance of the larger system will be verified to ensure that the exposure atmospheres match those agreed upon in the go/no go decision. Operating trials will be

conducted to optimize the stability of the system (reproducibility from day to day). Standard operating procedures will be developed.

Task 2: Conduct Animal Exposures

Rodents of species, strains, genders, and ages appropriate for each of the different health outcome assays will be exposed by inhalation 6 hours/day, 7 days/week for times ranging from a few days to 6 months to one of four dilutions of the exposure atmosphere or to clean air as negative controls. Although the longest exposure of any animal will be six months, not all animals will begin exposure at the same time. It will require approximately eight months of continuous exposure days to complete the entire protocol.

Task 3: Conduct Health Measurements and Analyze Data

Health evaluations will focus on effects in the respiratory and cardiovascular systems, although indicators of general systemic toxicity will also be measured. Measurements of the effects will be done at different times during the study, and after different lengths of exposure. General toxicity will be evaluated by clinical condition and body weight, hematology, blood chemistry, and blood clotting factors, cells and chemical mediators in bronchoalveolar lavage fluid, and histopathology of the respiratory tract after 7 days and 6 months of exposure. Alteration of the expression of genes in lung tissue will be determined after 7 days and 6 months of exposure. Because cancer is of concern but cannot be tested adequately without lifetime exposures, multiple indices of pre-cancer changes will be measured, including methylation and oxidative injury of lung DNA and formation of micronuclei (chromosome damage) after 7 days and 6 months of exposure. Resistance to sublethal respiratory infection with virus (Respiratory Syncytial Virus) and bacteria (Pseudomonas aeruginosa) will be determined after 7 days of exposure. The electrocardiogram will be monitored continually by telemetry for 3 days before exposure, during 7 days of exposure, and for 4 days after exposure, and heart and vessel tissues will be examined for histopathology. The effect of 3 days of exposure on amplification or suppression of respiratory allergic responses in animals made allergic in advance will be tested. The effect of exposure on the development of respiratory allergy will be tested by exposing animals *in utero* and for a short time after birth and then testing their susceptibility to sensitization to an allergen.

The biological response data will undergo well-established, rigorous quality control and quality assurance reviews, including independent audits. Audited data will be entered into an existing standardized database, and only those data will be analyzed and reported as final results. Exposure-response relationships will be tested using established procedures involving both trend tests and comparisons to control. The nature (shape) of the dose (exposure)-response relationships will be determined, including thresholds for significant responses (to the extent thresholds can be estimated from four discrete exposure levels). The effects of exposure to coal emissions will be compared to those resulting from exposure to other source emissions

Task 4: Characterize the Exposure Atmospheres and Analyze Data

The composition of the exposure atmospheres at all exposure levels (and the control atmosphere) will be determined in extensive detail. Some parameters will be measured in real time or daily to monitor and control the exposures. Detailed speciation of the atmospheres will occur at three times during the exposure period. Particle mass and number size distributions and particle size-specific composition will be determined. Pollutant gases will be measured. Both volatile and semi-volatile organic gases and vapors will be speciated in detail, and the gas-particle distribution of semi-volatiles will be determined.

The exposure characterization (composition) data will undergo well-established, rigorous quality control and quality assurance reviews, including independent audits. Audited data will be entered into an existing standardized database, and only those data will be summarized and reported as final results.

C. DELIVERABLES

Quarterly, topical, and final reports will be submitted in accordance with the Federal Assistance Reporting Checklist and other terms that may be established in the specific funding agreement for this project. The recipient will also send informal, monthly updates on project status to the COR via email. A report summarizing the entire results of Phase I and proposing a strategy for the exposures under Phase II will be submitted regardless of other requirements. This report will serve as a basis for the go/no go decision on beginning Phase II.

The primary goal for communicating the results of this project will be the publication of results in peer-reviewed scientific literature. A minimum of one publication will describe the experience during Phase I, including coal preparation, the operation of the furnace/emissions modification/exposure chamber system, and the atmospheres generated using the two coal types. Publications resulting from Phase II will include one describing the exposure atmospheres in detail and multiple papers describing the results of various health assays. It is anticipated that a separate paper will focus on comparing health responses to the coal emission exposure to the responses to exposures to the other source emissions studied using the same protocol up to that time (i.e., diesel and gasoline emissions, hardwood smoke, and street dust).

D. BRIEFINGS/TECHNICAL PRESENTATIONS

Briefings will be given at the sponsor's location as requested. It is anticipated that at a minimum, a "kickoff" briefing will be given soon after the award, another to discuss the results of Phase I, and at least one other to discuss the results of Phase II. Technical presentations and papers will be given as directed at DOE/NETL Annual Contractor's Review Meetings.

Technical presentations describing methods, interim results, and final results will also be made at scientific meetings appropriate for the topics and investigators as the project progresses. Examples of candidate meetings include Air and Waste Management Association, American Association of Aerosol Research, Society of Toxicology, American Thoracic Society, and specialty topical meetings and conferences.

In addition, progress will be reviewed together with progress on results from exposures to other source emissions at the annual meeting of the NERC External Scientific Advisory Committee and sponsors, which will include DOE/NETL representatives. DOE/NETL managers will also be included in all communications to sponsors pertaining to the NERC program.